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论著

曲尼斯特延缓糖尿病肾病肾间质纤维化的作用及机制

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摘要:

目的：研究曲尼斯特延缓糖尿病肾病肾间质纤维化的作用及机制。方法：建立糖尿病肾病（DKD）大鼠模型：SD大鼠随机分为正常对照组（*n*=6）、DKD模型组（*n*=8）、曲尼斯特低剂量（*n*=8）和高剂量治疗组（*n*=8）。采用高糖高脂饲料喂养联合低剂量STZ注射构建大鼠DKD模型。成模后，分别予以曲尼斯特200 mg/（kg·d）（曲尼斯特低剂量组）和400 mg/（kg·d）（曲尼斯特高剂量组）分2次灌胃。于第8周末处死大鼠，收集大鼠24 h尿液测24 h尿白蛋白排泄量，收集血测肾功能及血白蛋白；取部分肾组织置于4%中性甲醛溶液中固定，采用免疫组织化学检测肾组织补体C3a受体（C3aR），E-钙黏附蛋白（epithelial cadherin，E-Cadherin），*α*-SMA，纤维连接蛋白（fibronectin，FN），I型胶原蛋白（collagenI，Col I），干细胞生长因子（stem cell factor， SCF）和干细胞因子受体（c-kit）的表达以及分布；Western印迹检测肾组织E-cadherin，*α*-SMA，FN，Col I，SCF和c-kit蛋白的表达；RT-PCR检测肾组织FN，Col I，SCF，c-kit mRNA的表达。结果：曲尼斯特能抑制肥大细胞在DKD大鼠肾组织的浸润；DKD模型组肾小管上皮细胞E-cadherin的表达较正常对照组减少，并可见*α*-SMA表达，曲尼斯特可一定程度逆转这一过程；与正常对照组比较，DKD模型组肾小管间质区域Col I和FN的表达增加，曲尼斯特能剂量依赖性地抑制Col I和FN的表达；DKD大鼠肾组织SCF，c-kit蛋白及mRNA表达增加；肾组织SCF，c-kit蛋白表达与肥大细胞浸润程度及肾小管间质FN，Col I蛋白的表达呈显著正相关。曲尼斯特能抑制SCF，c-kit mRNA及蛋白的表达（*P*<0.05）。结论：肥大细胞参与了DKD大鼠肾间质纤维化的发生发展，曲尼斯特可能通过阻断SCF/c-kit信号通路，抑制肥大细胞的募集而逆转DKD大鼠肾小管上皮细胞的EMT，抑制肾间质纤维化。

关键词： 曲尼斯特 糖尿病肾病 肾间质纤维化 肥大细胞 干细胞生长因子 干细胞因子受体

Role and mechanism of tranilast preventing the progression of tubulointerstilial fibrosis in diabetic kidney diseases

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Abstract:

Objective: To determine the role and mechanism of tranilast preventing the progression of tubulointerstilial fibrosis in diabetic kidney disease (DKD). Methods: Sprague-Dawley rats were randomly divided into a control group (*n*=6), DKD model group (*n*=8), low dose tranilast group [200 mg/(kg.d), *n*=8], and high dose tranilast group [400 mg/(kg.d), *n*=8]. Tranilast was administered daily after the model was built. Rats were sacrificed at day 56, 24 hour urine was collected to measure 24-hour urine albumin excretion, and blood was collected to determine the renal function and serum albumin. Then the kidneys were harvested and subjected to studies. The expression of C3aR, E-cadherin, *α*-SMA, fibronectin(FN), collagen I (Col I), stem cell factor (SCF) and c-kit were detected by immunohistochemical staining respectively. The expression of E-cadherin, *α*-SMA, FN, Col I, SCF and c-kit protein was analyzed by Western blot, and the expression of FN, Col I, SCF and c-kit mRNA was examined by RT-PCR. Results: Tranilast can inhibit the infiltration of mast cells in the kidneys of DKD rats. The expression of *α*-SMA in the kidneys of DKD rats inereased significantly (*P*<0.05), while the expression of E-cadherin decreased (*P*<0.05). Tranilast increased the expression of E-cadherin and decreased the expression of *α*-SMA in the prophase of DKD dose dependently. The expressions of FN and Col I were increased in the tubulointerstitial fields in DKD model rats (*P*<0.05). After the tranilast treatment, these changes were relieved to a certein degree (*P*<0.05). The expression of SCF and c-kit in the tubular and interstitial tissue was slight. The increased expressions of SCF and c-kit protein and mRNA in DKD model rats were downregulated by tranilat (*P*<0.05). The expressions of SCF and c-kit were positively correlated with the infiltration degree of mast cells and the expressions of FN, Col I. Conclusion: Mast cells participate in and aggravate the renal tubulointerstitial fibrosis in DKD rats. Tranilast can reverse the EMT of renal tubular cells and inhibit the tubulointersitial fibrosis of DKD by blocking the infiltration of mast cells induced by SCF/c-kit pathway.

Keywords: tranilast diabetic kidney disease tubulointerstitial fibrosis mast cells stem cell factor c-kit
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